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ACUTE LACUNAR DIPHTHERIA OF THE TONSILS,

WITH STUDIES ON THE RELATION OF THE REAL TO THE PSEUDO *BACILLUS DIPHTHERIÆ.*

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The class of cases forming the subject of this paper are those in which diphtheria either begins or runs its entire course with the local picture of a lacunar or follicular amygdalitis. These cases, whose clinical study has been the subject of much difference of opinion, are becoming more important daily. It is through the avenue of these cases that diphtheria, sometimes of a fatal character, is unsuspectingly spread broadcast. If we are to advance in the prophylaxis of diphtheria we must do this not through means of the pronounced cases, in which the membrane is apparent even to the lay observer, but through the cases which in every way simulate the ordinary "sore throat," or tonsillitis, or lacunar or follicular amygdalitis. In my first paper upon diphtheria published in this journal I made a study of the *doubtful cases* of diphtheria. In those cases the diphtheria began or persisted as a simple angina or was manifest in the form of minute doubtful specks of ex-

presented by the author

udate, "punctate membrane" upon the tonsils or soft palate, or was found to begin or run its course as an amygdalitis, or, beginning as a lacunar amygdalitis, would terminate in a descending diphtheria with stenotic symptoms. For the sake of unity the writer has confined himself in the present paper to the study only of those cases of diphtheria which begin or run their entire course with the *clinical picture* of a typical lacunar amygdalitis.

Anatomically the assumption of a form of diphtheria which may present a lacunar distribution can be more easily understood if we study what transpires in the normal tonsil as well as in the inflamed structure. Stöhr proved conclusively that in the tonsils there is normally a constant diapedesis of leucocytes from the central follicles of the organ through the several layers of flat epithelium on the surface of the tonsil. These leucocytes reach the surface of the tonsil and are found in small masses between the epithelial cells of the external layer of the tonsil. They are thrown thus into the cavity of the mouth and are found also in the lumen of the lacunæ.

Heubner, studying the abnormal conditions that are found in diphtheria, states that this normal lymph current, if we may so call it, is replaced by an abnormal exudate of lymph and fibrin from the vessels in the interior of the tonsil. This fluid is exuded through the several layers of surface epithelium, and when it reaches the surface of the tonsil it coagulates. Its youngest form is in the shape of a coagulated network of fibrinous exudate, containing in its meshes the flattened horny epithelium of the uppermost layer on the tonsil. Thus for the first few days in the history of diphtheria we may have this coagulated exudate manifest itself in streaks or spots on the surface of the tonsil, indicating the situation of the lacunæ, follicles, or crypts, and the irregularities of surface between these

structures. It is foreign to the purpose of this study to enter the pathological field dealing with the structure of the forms of diphtheritic exudate or membrane.

Modes of Investigation.—The cases studied in this paper have been investigated both from a clinical and bacteriological standpoint. As to the latter mode of study, the methods pursued in all cases were exactly those utilized in the first paper of the writer which appeared in this journal August 27, 1892. Special stress is laid upon the animal experiment as the only positive proof of the virulence of the Loeffler *Bacillus diphtheriæ*. From the ordinary crude clinical, diagnostic, or hygienic standpoint it can be said that an adept can easily identify the Loeffler bacillus by its form, lack of motion, growth on media, and bouillon reaction (Roux, Yersin, Escherich), without going through the tedious animal tests. Yet this method, moderately certain for rapid diagnosis, is scarcely available for permanent and reliable scientific work.

The cases in this paper have thus been subjected to the above mentioned test and the number is limited. After proving certain theses the further accumulation of material has no additional scientific value.

Clinical Course.—The cases collected in this study divide themselves quite distinctly into three leading groups.

The first group of cases are those whose course is an exceedingly mild one—so much so that the patients show little or no constitutional disturbance. The appetite is good; the children are up and about; they complain of slight uneasiness in the throat; sometimes these throat symptoms are absent. Examination of the fauces shows a general hyperæmia; the tonsils are enlarged. When the patient gags, the tonsils become still more prominent and reveal the enlarged open lacunæ. There is absolutely no membrane

to be seen on the surface of the tonsil, but there is a yellow spot here and there which, on investigation, will be shown to be the opening of a lacuna or follicle or crypt. The lymph nodes at the angle of the jaw may or may not be enlarged. The above-described local symptoms are so mild that they may have improved or even disappeared the following day. The patient then is to all intents recovered from this so-called attack. We will show the fallaciousness of this assumption in our cases.

Case I.—Boy, aged six years, has been ill only for twenty-four hours; complains of soreness in his throat. Inspection shows the lacunæ filled with a fibrinous material on the right side, which can be scooped out of the tonsillar lacuna without difficulty. There is on the surface at the anterior border of the tonsil a minute fibrinous streak. Lymph nodes at the angle of the jaw on the right side enlarged. Temperature 101·8° in the axilla; the boy's condition is excellent and he does not feel ill.

Bacteriological examination of the fibrinous plug removed from the lacuna of the right tonsil and spread upon blood serum and subsequently isolated shows Loeffler bacilli virulent to guinea-pigs in dosage of 0·5 c. c. of bouillon culture within forty-eight hours. On the following day the tonsils still large, but the fibrinous streaks of exudate had disappeared.

A week subsequent to the above, the boy had completely recovered and the tonsils had returned almost to the normal size and appearances; they were still a little large and slightly red. A minute scoop was entered into the lacuna of the right tonsil as near as possible in the same situation as the first plug. This tonsil had this lacuna near the anterior border.

Diphtheria bacilli found virulent in same dosage as above.

Two weeks after the onset of the disease a similar plug removed from the same tonsil in the same locality.

Diphtheria bacilli still virulent in dose of 0·5 c. c. of bouillon culture forty-eight hours old.

At this time the appearance of the fauces was absolutely normal; no lacunar plugs to be seen. The above fibrin plug

was extracted from the depths of the lacuna of the right tonsil. The case is continued in Part II of this paper.

Case II.—Sister of the preceding, aged eleven years, had absolutely no subjective symptoms yet tonsils were enlarged and swollen, lacunæ open and wide, no elevation of temperature; child denies even soreness of the throat or any feeling of illness. Examination of the contents of the lacunæ scooped out as above shows what to all appearance were the Loeffler bacilli. They were not tested on animals.

A week subsequent to the first visit these bacilli had disappeared.

Case III —Minnie B., aged eleven years. A baby sister of patient had been ill two months previous with what, from the history, might have been diphtheria. The mother, who from this time inspected the throats of the children daily, found on the day of the visit to the writer slight redness in the throat.

Status —Tonsils are swollen and red; there is no membrane; the lacunæ contain a soft yellow substance which can be removed in form of a plug (fibrinous). The whole picture one of lacunar amygdalitis. Temperature is slight, so that it was not taken. No glandular enlargements. Child in the best of spirits and condition. Examination of the plug, removed with sterilized scoop from the lacuna, showed diphtheria or Loeffler bacilli virulent in bouillon culture forty-eight hours old in dose of 0·5 c. c. to guinea-pigs.

Two weeks subsequently the angry appearance of the tonsils had disappeared. The one lacuna from which a yellow plug had been removed no longer showed reaccumulation of exudate. Child protests she is not ill and wants to return to school, which, of course, was not permitted.

Two weeks after the first examination a plug was removed from the lacuna in the same tonsil in about the same situation as the first plug. The bacilli in this plug were no longer virulent. They will be discussed under the heading of the pseudo bacillus.

In the above-cited cases we have a very mild form of tonsillar lacunar diphtheria—mild from the outset, watched

all along its course, absolutely no membrane anywhere, and the children at no time during their illness in bed. The sterilized scoop, entered into the same tonsils at the same situation, showed that, as in the first case, there were bacilli in the depths of the lacunæ fully up to three weeks after the boy had shown no signs of lacunar plugs pouting from his tonsils. His case will be continued under the pseudo heading, and it will be shown that, as in Case III, the virulent was finally replaced by a non-virulent bacillus. Roux and Yersin, in investigations upon membranous diphtheria of the tonsils, recorded cases in which bacilli of Loeffler had persisted for fully five weeks after the disappearance of membrane. The foregoing, I believe, are unique as showing the persistence of virulent bacilli in the depths of the tonsillar lacunæ for weeks after complete convalescence. The non-virulent bacilli found in Cases I and III will be discussed in the second part of this paper.

The second group of cases of lacunar diphtheria are those in which the symptoms, both local and constitutional, re s omewhat more severe than in the first group of cases. The patients are quite ill; they suffer as do those patients who are affected with a severe follicular or lacunar amygdalitis of a simple infectious non-specific type (streptococcus). The fever is quite high, the prostration is more marked, and there are other symptoms of constitutional disturbance, such as pains in the limbs, furred tongue, and loss of appetite. Locally the tonsils are seen to be enlarged, red, and there exudes from the lacunæ a soft yellow fibrinous exudate. There is no membrane on the surface of the tonsil. There are glandular or lymph-node enlargements at the angle of the jaw. The following will illustrate:

Case IV.—Annie C., aged four years, has been ill one day. She complains of sore throat; tonsils are quite large and red; they present the typical appearance of a lacunar amygdalitis.

When the patient gags, the tonsils protrude pink-red in color and from the lacunæ there is expressed by the act of gagging a soft, yellow, consistent material. Temperature 101°, axillary. Child shows some prostration, but general condition is good Lymph nodes at the angle of the jaw on the left side can just be felt.

Examination of the lacunar contents as exuded shows Loeffler bacilli in almost pure culture, virulent.

Case V.—In this case the amygdalitis was complicated by symptoms of laryngeal involvement. These cases are important, showing as they do that, no matter how typical the lacunar amygdalitis, the larynx may be affected not by a simple but by a diphtheritic process, as are the tonsils themselves. These cases are especially apt to be judged as simple and non-specific.

Case VI.—Male, aged five years, has been coughing for three days. The tonsils are enlarged and have the typical follicular appearance. When the boy gagged it seemed as if the uvula, which curved upward, showed a minute yellow speck at its point. The voice is harsh and the cough croupy. No laryngeal breathing. Prostration marked. Bacteriological examination of the lacunar contents showed Loeffler bacilli.

The third group of cases of lacunar diphtheria are those which from the outset show a malignant course. They do not remain of the pure lacunar type after the first few days. In the following case the change occurred on the third day. The nasal mucous membrane is drawn into the symptomatic picture of the disease. We thus have a case beginning in every way like a severe follicular amygdalitis of a non-specific type (streptococcus), ending in a fatal diphtheria with pronounced septic diphtheritic phenomena.

Case VII.—Baby, female, aged thirteen months. There had been a number of cases of "sore" throat in the family (seven children), especially in that of a boy of eight years, whose throat, I regret to say, I did not examine bacteriologically. There was no membrane in this boy's throat, and there was every appearance when I saw him of a simple non-specific an-

gina. The mother also had suffered from a "sore" throat. The symptoms in all cases had been so mild as not to call for any outside advice. The baby had been ill for two days when first seen. The infant was very pale and quite prostrated; rectal temperature, 102°. The lymph nodes on both sides of the neck, at the angle of the jaw, were much enlarged in packets. There was no skin eruption. The tonsils were enlarged, red, eroded, but had the appearance of a very severe lacunar amygdalitis; no nasal discharge. The following day the tonsils appeared less swollen and the amygdalitis an ordinary one. In view of the constitutional symptoms and a slight nasal discharge, I warned the mother to allow me to see the patient again if improvement was not rapid. Two days subsequent to this the tonsils had again swollen; the discharge from the nose became much more pronounced. The tonsils were so much swollen as not to permit a view of the posterior pharynx. The discharge from between the tonsils now contained minute shreds of what proved to be membrane.

Bacteriological examination of these shreds show Loeffler bacilli and large numbers of Roux's cocci. The case ended fatally, with symptoms of septic diphtheria, including extensive glandular infection.

The Diagnosis.—The constitutional symptoms give us very little clew to our diagnosis in the first group of cases, whereas in the third group the severe constitutional prostration from the outset leads us to suspect something more specific than simple amygdalitis.

The range of *febrile* disturbance is also quite unreliable. *Glandular swelling* may be practically absent in the mild cases, though it will be seen that in some of these cases we had as an accompaniment of the diphtheria a swelling of the lymph nodes on one side only. On the other hand, glandular swelling may be very marked in non-diphtheritic amygdalitis (streptococcus). The swelling of nodes on one side is peculiar to diphtheria, though I have very often

seen it in non-diphtheritic cases. *Albuminuria* may be present or absent at first, when diagnosis is most important. It is apt to be present in simple streptococcus disease as well as in the diphtheritic cases.

Henoch says, in a recent article upon the early clinical diagnosis of diphtheritic amygdalitis, that we must judge our cases from the consideration of a completed picture of the disease. It will also be seen that the diagnostic elements presented by this great clinician—the fibrinous exudate from the lacunæ, the confluence of patches of fibrin on the surface of the tonsil, affection of both tonsils, the uvula and nasal mucous membrane, albuminuria, the occurrence of several cases in one family—all apply either to the severe cases (third group), or appear late in the disease. To the writer it still seems that the only reliable test is the bacteriological one. There is now such a reaction as to encourage the hope that soon this test will be applied by the general practitioner in daily practice.

Part II.—Bacteriological.

The methods pursued in the investigation of the cases in this paper included not only an examination of the secretions on the surface of the affected tonsils, but what seemed more interesting and, in fact, imperative was the examination of the contents of the individual lacunæ of the tonsils. The lacunæ could only be studied in the older children, where they are of some size and in which it is possible to introduce a minute sterilized scoop or harpoon. In this way the plug or secretion in the depths of the lacunæ was obtained for study, spread upon serum, and isolated finally upon agar-agar. The writer has been impressed by the fact that the tonsil may harbor for weeks in the depths of its lacunæ, follicles, or crypts, as we may choose to call these structures, virulent diphtheria bacilli. In membranous

diphtheria we are not surprised to find, after the membrane has cleared away, that the diphtheria bacilli persist in the numerous ulcerations or erosions left by the membrane on the surface. In lacunar diphtheria, as in Cases I and III, where no membrane is formed in the whole course of the affection, we are surprised to find in the depths of the lacunæ fibrinous plugs which persist for weeks and which contain myriads of virulent bacilli. Roux and Yersin found that in cases of *membranous* diphtheria the *Bacillus diphtheriæ* persisted in a virulent state as long as five weeks after all traces of membrane had disappeared from the fauces. I have had this condition of things repeated in lacunar diphtheria. The *virulence* of the *Bacillus diphtheriæ* can be preserved for an exceedingly long time outside of the body. It resists the drying process if shielded from the direct sunlight. A culture of the *Bacillus diphtheriæ* in the author's possession, after having dried for months in an ordinary test-tube, so that the original agar was reduced to a dry crisp, showed after nine months a virulence in bouillon forty-eight hours old to the amount of half a cubic centimetre to old or young guinea-pigs. The scrapings of such a culture, though containing many involution forms such as have been pictured by the writer in his first article, grew most luxuriantly in agar and bouillon. Old bouillon cultures, reduced by age from evaporation to a few drops from ten cubic centimetres, were found to abound in virulent living bacilli which grew upon agar. Roux and Yersin, in their last article upon diphtheria, assume that the mild cases may yield bacilli which show a diminished virulence in animal experiment. The writer's experience, at least in the class of cases examined, does not tend to confirm this view. Bacilli from the very mildest cases of lacunar diphtheria were found in pure culture virulent to guinea-pigs in dosage of half a cubic centimetre of bouillon culture forty-eight hours

old. This dose is now conceded by most writers to be small enough as a test of virulence. All animals injected with such an amount died within forty-eight hours with symptoms of virulent diphtheria (local hæmorrhagic œdema, enlarged lymph nodes, pleuritic and peritoneal effusion).

In Case I the Loeffler bacillus was isolated three weeks after all traces of tonsillar trouble had disappeared. These bacilli were preserved in separate cultures pure, and those isolated latest were found fully as virulent as the bacilli isolated at the outset of the disease. All these bacilli were cultivated from the lacunar plugs so often mentioned in this paper. A dose of half a cubic centimetre of a bouillon culture invariably proved lethal to guinea-pigs of medium weight (three hundred and fifty to four hundred and fifty grammes) within forty-eight hours. In this way the bacilli obtained from the mild cases were compared with a very old culture which has been in the author's possession over two years and which has been isolated by him from a virulent membranous diphtheria. Of this culture a tenth of a cubic centimetre will prove lethal to animals in varying time, but half a cubic centimetre will invariably prove lethal in forty-eight hours if bouillon cultures forty-eight hours old are used. In short, bacilli of a so-called diminished virulence—those which would require two or three cubic centimetres to prove lethal from bouillon cultures forty-eight hours old—were sought by the writer in the mild cases, not of membranous but lacunar diphtheria, and were not found. In the author's experience, the mildest cases furnish as virulent bacilli as the most severe. In all this we assume that certain fixed laws in the preparation of all cultures are followed. The agar cultures are made from single colonies upon alkaline agar one per cent. These agar cultures are taken out of the thermostat after forty-eight hours. Bouil-

lon cultures of simple pepton alkaline bouillon are prepared from these agar tubes. The bouillon is taken out of the thermostat after forty-eight hours. The animals used in testing virulence are always *young animals*, three hundred and fifty to four hundred and fifty grammes in weight. The bouillon cultures should be eight to ten cubic centimetres in bulk and the supernatant fluid above the sediment should not be poured off, as has been done by many authors, and the thick residue used, but the whole culture is shaken up and half a cubic centimetre of this is used. Old animals even of medium weight show a certain amount of resistance to diphtheria and should not be used. The above are brought to the notice of the worker in diphtheria as a result of a long and tedious series of studies in this field. They will insure uniform results.

The Pseudo Bacillus Diphtheriæ (Hofmann, Loeffler).—In the first paper of the writer four cases were recorded in which a bacillus was found which closely resembled the real bacillus, reacted much the same upon culture media, but lacked virulence completely. In the studies of this paper the writer has succeeded in isolating a bacillus from cases in which the virulent diphtheria bacilli had been found and which appeared in the same patient after a lapse of weeks. In ordinary stained preparations (Loeffler blue) an expert would have great difficulty in finding marked differences of form. They were lacking completely in virulence.

The relationship of the real to the pseudo *Bacillus diphtheriæ* has lately been the subject of a critical review by Escherich. This critique was incited by the surprising discoveries of Fraenkel, who described a bacillus which in every way corresponded to the true Loeffler bacillus as to culture and stain, but which differed from it only in lacking virulence. Doses of two cubic centimetres seemed to be well borne by animals, whereas in five-cubic-centimetre

doses of a bouillon culture a lethal result followed. Fraenkel isolated his bacillus from the human conjunctiva in cases in no way related to diphtheria. He concluded from his observations that he had before him the true *Bacillus diphtheriæ*, but reduced in virulence; that the bacillus he had isolated and the Hofmann pseudo bacillus were identical. In this way he championed the cause of Roux and Yersin, who maintain the same ground. Escherich does not agree with these observers. The writer of the present paper is inclined to look upon the whole question of relationship of these two bacilli as an open question. But little has been proved on either side. The question is one of the widest range of importance. If the real *Bacillus diphtheriæ* can so completely lose its virulence as will be seen in the bacillus isolated in this study, certainly all known means have as yet failed to bring it back to its original state of virulence. Pasteur, in reducing the bacillus of charbon to a non virulent organism, did so by artificial means outside the animal economy. I therefore do not consider this fact brought forward by Roux and Yersin in support of their views as quite apt or all-satisfying.

The following studies are certainly unique and are published at this time as peculiarly pertinent to this very important question of the relationship of the real to the pseudo *Bacillus diphtheriæ*.

Cases in which a bacillus, non-virulent, replaced the real *Bacillus diphtheriæ* after a lapse of time:

Case I, *lacunar diphtheria*, virulent *Bacilli diphtheriæ* found up to three weeks after convalescence, then the non-virulent bacilli were found.

If the reader will refer to Case I of the first group of cases of lacunar diphtheria it will be seen that up to the third week after the onset of the illness, and fully that time after the boy felt well and the tonsils had cleared, the tonsils and

pharynx were in appearance normal, and were the seat of virulent bacilli.

Three weeks, therefore, after the onset of the illness a sterilized silver scoop was introduced into the lacuna of the right tonsil and a fibrinous yellow plug was removed. This plug was found to contain almost exclusively a bacillus in every possible way as to shape and stain the exact counterpart of the real *Bacillus diphtheriæ*. It was non-motile, stained with Loeffler blue in broken areas, and was not decolorized by Gram. There were clubbed shapes and involution forms in abundance. A very close study of the virulent bacillus taken from the same case and tonsil earlier in the disease (fourteen days after the onset) and this present bacillus side by side showed a very slight difference in thickness of the two bacilli; the non-virulent seemed to stain a little thicker, but so slight was this difference that unmarked there is for practical diagnostic purposes but very little to fix upon, for at times the virulent bacilli are thicker than others, short and long. The length of both bacilli seemed and still appears identical. In growth there are distinct differences.*

Bouillon is uniformly clouded after forty-eight hours and the deposit is much more abundant than that of the virulent bacillus. The reaction of the bouillon is distinctly alkaline after forty-eight hours.

Agar-agar yields a growth in which we at once notice a luxuriance foreign to the virulent bacillus. The colonies upon agar-agar reach a fuller development in shorter time than the real bacillus. They seem whiter and more opaque.

The above non-virulent bacilli were found up to the end

* Measurements of the Loeffler virulent bacillus: Length, 2·2 μ to 5 μ; width, 0·6 μ to 0·8 μ. Some non-virulent bacilli (pseudo) of same case: Length, 2·6 μ to 4 μ; width, 0·6 μ to 0·78 μ. Involution forms, of course, vary widely from above.

of the fifth week after the onset of the illness and were isolated in pure culture and are in the possession of the writer. The attempt to find them in the tonsil six weeks after the onset of the illness resulted only in their identification in smear preparations upon serum; they were overgrown in twenty-four hours by staphylococci.

Case II, in which the *virulent Bacillus diphtheriæ* is replaced in the same tonsil by a *non-virulent bacillus*.

This is Case III of the mild series of lacunar diphtheria cited in the first part of this paper (refer to history). The patient was seen four times, the second visit a day after the first. The tonsils had cleared, and the yellow streaks and lacunar appearances had disappeared; the tonsils were still red and enlarged. The *Bacillus diphtheriæ*, isolated from the right tonsil, had been found virulent.

Third visit a week after the first one, and the tonsils apparently normal in size and appearance: *no culture made.* Fourth visit, fifteen days after the first: tonsils normal; child well; a sterilized scoop entered into the lacuna of the right tonsil, behind the anterior faucial pillar, as near as possible to the place where the virulent bacilli had been extracted. A large, yellow plug, which had been invisible, was removed from the depths of this lacuna. From this plug there was obtained a luxuriant growth upon blood serum of a bacillus the exact counterpart of the non-virulent bacillus isolated from Case I above.

In the above two cases we have typical lacunar diphtheria in which no membrane was seen at any period of the disease, both of which cases were exceedingly mild in all their manifestations. In both cases the real bacillus—virulent—persisted for three weeks in one case and up to the second week in the other. The bacilli were all isolated from plugs of fibrinous exudate removed from the lacunæ and sown upon blood serum. The various sets of bacilli

were kept separated, and tested and found all to be of a like virulence, as stated in another part of this paper. At the beginning of the fourth week in one case after the first isolation of virulent bacilli, and the beginning of the third week in the other case, these virulent bacilli of diphtheria were replaced by a bacillus which was in every way, except in growth upon agar and in bouillon, the most accurate counterpart of the virulent original bacillus. It was, however, in both cases a harmless, non-virulent bacillus. Loeffler, in his second article upon diphtheria, asserts that in a case of an adult he had found the virulent and the non-virulent bacilli growing side by side upon the agar plate, and had isolated both. In this way he accounted for the famous pseudo *Bacillus diphtheriæ* of Hofmann, saying that the pseudo bacillus was an accompanying harmless bacillus of the real micro-organism. Roux and Yersin found the non-virulent or pseudo bacilli in cases of membranous diphtheria which early in the disease had yielded the virulent bacillus. The writer's cases are the only ones in which the real and virulent bacillus was replaced in the same tonsil and, as near as could be judged, in the same locality, entirely by a rich growth of a non-virulent bacillus which the expert examination alone could only find the least remove thicker than the virulent bacillus, but of the same form and staining peculiarities. In order to be explicit, we wish to repeat that this bacillus is non-motile, grows more luxuriantly upon agar than the virulent bacillus and the bouillon cultures (peptone bouillon) are not acid, as in the real bacillus, but alkaline after forty-eight hours in the thermostat. The bouillon deposits a more abundant sediment than the virulent and is more diffusely clouded. Roux, Yersin, and Escherich have called attention to the early acidity of the bouillon cultures of the virulent bacillus (forty-eight hours), and the author has had ample op-

portunity to verify this important and invariable quality of the virulent bacillus. The non virulent bacillus stains exactly like the virulent micro-organism both in Loeffler blue and Gram stains. As stated, it seems, when closely studied *side by side* with the virulent bacillus, broader than the latter. I doubt whether, in unmarked specimens, the writer's non-virulent bacilli could be distinguished by stain alone. We have the same clubbed and involuted forms shown by the virulent bacillus. I have noticed that it does not grow so luxuriantly in sugar bouillon as it does in the simple peptone bouillon. In the latter the shape of the non virulent bacillus is more complete as a bacillus form after forty-eight hours than that of the virulent bacillus, which is often seen in abortive forms at this time in bouillon culture.

Animals do not react even to large doses (eight cubic centimetres of forty-eight hours' bouillon) of the non-virulent bacillus; a few guinea-pigs develop a slight œdema, which eventually disappears. After having received these large quantities of the non-virulent bacillus, four guinea-pigs were reinjected after the lapse of a week with half a cubic centimetre of the virulent bacillus with lethal effect in two cases within forty-eight hours; a third, in three days; and a fourth, an old, fully-developed pig, in four days. Thus the non-virulent bacillus was not a vaccine against the virulent bacillus. These experiments, it will be recalled, were also performed with the non-virulent bacilli isolated in four cases of the first paper on this subject by the writer with similar results.

The literature of the non virulent or pseudo *Bacillus diphtheriæ* is still in its infancy. Hofmann, Loeffler, Escherich, Fraenkel, Welch, Abbott, and the writer of this article have studied it. Its rôle and exact place in the morphology of diphtheria are still a mystery. The German school of bacteriologists all unanimously agree as to its

place among the harmless saprophytes. Fraenkel is an exception to this view and sides with Roux, who thinks that the non-virulent pseudo bacillus is a weakened form of the virulent bacillus and, under conditions not yet discovered, as stated was the case in charbon, may become a dangerous, virulent micro-organism.

In closing these studies upon diphtheria the writer wishes to emphasize the fact that the tonsil is an organ which possesses a circulatory activity hitherto not attributed to it. It was formerly looked upon as an anatomical appendage whose purpose was problematic. Stöhr has demonstrated the constant migration of leucocytes from the interior of the tonsil (follicles) to the surface, and therefore into the lumen of the lacunæ. The writer is inclined to believe that future work may reveal that the tonsil in this functional activity exercises a destructive influence upon pathogenic bacteria which may be on the surface of the tonsil or in the lumen of the lacunæ. Blood lymph and serum possess inherent destructive powers upon the life of bacteria (Nuttall, Buchner, Prudden). Leucocytes also have the power of attracting within their substance bacteria. Both these conditions exist in the tonsils. If we examine the lacunar plugs in lacunar diphtheria we find leucocytes which seem to contain within them cocci and bacilli. It may be that the prolonged retention of the *Bacillus diphtheriæ* for weeks in the depths of the lacunæ under the intimate influence of the above-mentioned phenomena of lymph and leucocyte activity may not be without effect on one phase of the vitality of the *Bacillus diphtheriæ*—its virulence. Such a theory as the above is offered with the greatest of diffidence in view of our limited knowledge of the actual relation of these two bacilli.

Note.—The experimental work of this paper was carried on in the Carnegie Laboratory.

Full List of Cases examined for Lacunar Diphtheria, and which all presented the Typical Clinical Picture of an Acute Lacunar or Follicular Amygdalitis.

Case No.	Sex.	Age.	Glandular enlargements at angle of jaw.	Bacteriological results.	Animal experiments.
1	Male.	6 yrs.	Right.	Loeffler bacillus.	Virulent.
2	Female.	11 yrs.	None.	" "	Not tested.
3	"	11 yrs.	"	" "	Virulent.
4	"	4 yrs.	Left.	" "	"
5	Male.	5 yrs.	Both.	" "	"
6	Female.	4 yrs.	None.	Streptococcus.	
7	Male.	3 yrs.	"	"	
8	"	1½ yr.	Right.	Loeffler bacillus.	Virulent.
9	"	13 mos.	.....	Streptococcus.	
10	Female.	4 yrs.	.....	Pseudo bacillus.	Not virulent.
11	"	20 mos.	None.	"	" "
12	"	2 yrs.	"	Streptococcus.	
13	"	1 yr.	"	Roux's coccus.	
14	Male.	2½ yrs.	"	Pseudo bacillus.	Not virulent.
15	"	3 yrs.	Both sides.	Bacilli not pseudo.	" "
16	"	4 yrs.	Right.	Loeffler bacillus.	Virulent.
17	"	3½ yrs.	Both sides slight.	Bacilli not pseudo.	Not virulent.
18	Female.	22 mos.	None.	Bacilli not pseudo, streptococci.	" "
19	Male.	8 yrs.	Both.	Streptococci, staphylococci, Roux's coccus.	
20	Female.	1 yr.	"	Streptococcus.	
21	Male.	1½ yr.	None.	Roux's coccus.	
22	"	15 mos.	"	Loeffler bacillus.	Virulent.
23	"	11 mos.	"	Roux's coccus.	
24	"	2 yrs.	Right.	Loeffler bacillus, Roux's coccus.	Virulent.
25*	Female.	13 mos.	Both.	Loeffler bacillus.	" "
26	"	15 mos.	Left.	Streptococcus, staphylococcus.	
27	Male.	5 yrs.	Both.	Streptococci, staphylococci.	
28	Female.	4 yrs.	Both.	Streptococci, Roux's coccus.	
29	Male.	3½ yrs.	.....	Streptococcus.	

* Marked Case V in paper.

Case No.	Sex.	Age.	Glandular enlargements at angle of jaw.	Bacteriological results.	Animal experiments.
30	Female.	3 yrs.	.....	Streptococcus.	
31*	"	2½ yrs.	None.	Loeffler bacillus.	Lost; not tried on animals.
32	"	11 mos.	"	Streptococci.	
33	"	3 yrs.	.....	Loeffler bacillus.	Virulent.
34	Male.	5 yrs.	.....	Streptococcus, Roux's coccus.	
35	Female.	2½ yrs.	Both.	Streptococcus, Roux's coccus.	
36	Male.	6 yrs.	None.	Streptococci.	
37	"	17 mos.	"	"	
38	"	4½ yrs.	"	Staphylococci, streptococci.	
39	"	6 yrs.	Left.	Staphylococci, streptococci.	

* Sister to patient in Case I in paper.

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